DATA EVALUATION RECORD

ALGAE EC_{50} TEST

GUIDELINE 123-2/840.5400 (TIER II)

1. CHEMICAL: 1,2-benzenedicarboxaldehyde (ortho-phthalaldehyde, OPA)
   PC Code No.: 129017

2. TEST MATERIAL: Ucarcide® P200 Purity: 98%

3. CITATION

Authors: Roshon, R.

Title: Ortho-phthalaldehyde: Growth Inhibition Test with the Freshwater Green Alga, *Selanastrum capricornutum* Printz

Study Completion Date: May 16, 2002

Laboratory: ESG International, Inc., Guelph, Ontario, Canada

Sponsor: The Dow Chemical Company, Midland, MI

Laboratory Report ID: S2041-01

DP Barcode: D285717

MRID No.: 457024-01

4. REVIEWED BY: Kathryn Montague, Biologist, RASSB, AD
   Signature: Date:

5. APPROVED BY: Norm Cook, Branch Chief, RASSB, AD
   Signature: Date:

6. STUDY PARAMETERS

Scientific Name of Test Organism: *Selanastrum capricornutum*

Definitive Test Duration: 96 hours

Type of Concentrations: Time-weighted average

7. CONCLUSIONS:

Results Synopsis (using time-weighted average concentrations):

Cell Number 96-hour EC_{50}: 0.222 ppm ai 95% C.I.: ppm ai

NOAEC: 0.101 ppm ai Slope:

Growth Rate 96-hour EC_{50}: 0.275 ppm ai 95% C.I.:ppm ai

NOAEC: 0.101 ppm ai

8. ADEQUACY OF THE STUDY

A. **Classification:** Core

B. **Rationale:** N/A

C. **Repairability:** N/A

9. GUIDELINE DEVIATIONS

1. Light intensity used during testing (8 +/- 20% kLux) was higher than recommended (4.3 +/- 15% kLux)

2. The level of detection (LOD) for OPA in this test was 0.2 mg/L. Therefore, only the stock solution and the three highest concentrations were analyzed. The lower concentrations were extrapolated from the measured concentrations of the higher levels. All concentrations were then time-weighted for use in determining the toxicity endpoints.

10. SUBMISSION PURPOSE: Study was submitted under FIFRA section 6(a)(2). The chemical tested is used as a materials preservative.

11. MATERIALS AND METHODS

A. Test Organisms

Guideline Criteria	Reported Information
Species *Skeletonema costatum* *Anabaena flos-aquae* *Selenastrum capricornutum* *Navicula pelliculosa*	*Selenastrum capricornutum*
Initial Number of Cells 3,000 - 10,000 cells/ml	10,000 cells/ml
Nutrients Standard formula, e.g. 20XAAP	algal growth medium (Env. Canada, 1992)

B. Test System

Guideline Criteria	Reported Information
Solvent	
Temperature	24 ± 1°C

Guideline Criteria	Reported Information
Skeletonema: 20°C Others: 24-25°C	
Light Intensity Anabaena: 2.2 K lux (±15%) Others: 4.3 K lux (±15%)	4 ± 10% k Lux for culturing; 8 ± 20% kLux for testing
Photoperiod Skeletonema: 14 h light, 10 h dark or 16 h light, 8 h dark Others: Continuous	Continuous
pH Skeletonema: approx. 8.0 Others: approx. 7.5	7.5 ± 1

C. Test Design

Guideline Criteria	Reported Information
Dose range 2X or 3X progression	2.5X progression
Doses at least 5	6 plus controls
Controls negative and/or solvent	negative

Guideline Criteria	Reported Information
Replicates per dose 3 or more (4 or more for Navicula)	5 per dose
Duration of test 120 hours	96 hours
Daily observations were made?	Yes
Method of Observations	Cellular counts with hemocytometer
Maximum Labeled Rate	n/a (materials preservative)

12. REPORTED RESULTS

Guideline Criteria	Reported Information
Quality assurance and GLP compliance statements were included in the report?	Yes
Initial and 96 h cell densities were measured?	Yes
Control cell count at 96 hr ≥2X initial count?	Yes
Initial chemical concentrations measured? (Optional)	Yes
Raw data included?	Yes

Results - Cell count

Nominal Concentration (mg ai/L)	Time-weighted average concentration (mg ai/L)	96-hr mean Cell Density (x 10^4 cells/ml)	% Inhibition	96-Hour pH
Control	0.000	329.4	---	7.51
0.01024	0.058	336.0	-1.98	7.56
0.0256	0.070	294.4	10.64	7.52
0.064	0.101	311.8	5.37	7.58
0.16	0.201	214.6	34.85	7.58
0.4	0.312	6.5	98.03	7.35
1.0	0.859	1.8	99.47	7.29

Results - Growth Rate

Nominal Concentration (mg ai/L)	Time-weighted average concentration (mg ai/L)	0 - 72h mean growth rate	0 - 72 h % Inhibition	72 - 96h mean growth rate	72 - 96h % Inhibition
Control	0.000	0.06939	---	0.06166	---
0.01024	0.058	0.06942	-0.04	0.06172	-0.10
0.0256	0.070	0.06588	5.06	0.06044	1.97

0.064	0.101	0.06328	8.81	0.06108	0.93
0.16	0.201	0.05502	20.71	0.05715	7.30
0.4	0.312	0.02323	66.52	0.02024	67.17
1.0	0.859	0.00865	87.54	0.00611	90.10

Statistical Results - using time-weighted concentrations

Statistical Method: Linear Interpolation for EC50; TOXSTAT - square-root transformation, followed by ANOVA with Bonferroni's test for 96-h cell count NOEC; ANOVA with Dunnett's Test for 96-h growth rate NOEC

Cell number 96-hr EC_{50}: 0.222 mg ai/L 95% C.I.: n/r

Slope: n/r NOAEC: 0.101 mg ai/L

72-96 hour growth rate EC50: 0.275 mg ai/L 95% C.I.:n/r

slope: n/r NOAEC: 0.101 mg ai/L

13. **Verification of Statistical Results** - using time-weighted concentrations

Statistical Method: Moving average analysis for EC50 (TOXANAL), for NOEC/LOEC (TOXSTAT)

Cell number 96-h EC_{50}: 0.184 mg ai/L 95% C.I.: 0.171 - 0.200 mg ai/L

Slope:4.38 (probit–not reliable for this data)	NOAEC: 0.101 mg ai/L
72-96 hour growth rate EC50:0.323 mg ai/L	95% C.I.: 0.294 - 0.353 mg ai/L
slope:3.80 (probit - not reliable for this data)	NOAEC: 0.101 mg ai/L

14. REVIEWER'S COMMENTS: The level of detection (LOD) for OPA in this test was 0.2 mg/L. Therefore, only the stock solution and the three highest concentrations were analyzed. The lower concentrations were extrapolated from the measured concentrations of the higher levels. All concentrations were then time-weighted for use in determining the toxicity endpoints.

Sign-off Date : 01/09/03
DP Barcode No. : D285717 and D285716